# Effect of Adding Granul Basil (*Ocimum americanum*) as Antioxidants in Fried Foods

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**Abstract**

Basil is one of medicinal plants in Indonesia that has been used empirically as antimicrobes, analgetic, antiinflammatory, antivirus, antitumor, and antioxidants that prevented ischemia. This research aimed to investigate the effect of adding granule ethanol extract of basil leaves (*Ocimum americanum*) as antioxidant to capture free radicals in fried foods. The antioxidant activity of ethanol extract of basil leaves with DPPH determinated by $IC_{50}$, comparing the antioxidant activity of the granule and the control by determination of quality properties of oil and food after frying using Fourier Transfor Infra Red (FTIR) spectra, and quantitative parameters by determination percentage of free fatty acid (FFA). Results of maceration with ethanol 15.94% w/w, total ash content 9.85%, water soluble ash content 3.98%, acid insoluble ash content 3.98%, water soluble extract 25.89%, ethanol soluble extract 12.76%, water content 6.87%, drying shrinkage 11.19%. Testing the antioxidant activity of the ethanol extract of basil leaves with DPPH gave $IC_{50}$ of 80.55 ppm. The result of antioxidant testing by determination percentage of FFA statistically with ANOVA (significance 0.00<5) and comparison of the concentration extract in basil granule with LSD test significantly different. The result indicates that adding ethanol extract of granules basil on fried foods gave antioxidant activity because it can inhibit the increasing of FFA percentage.

**Keywords**: Antioxidant, basil leaves, DPPH, granule

# Pengaruh Penambahan Granul Kemangi (*Ocimum americanum*) sebagai Antioksidan pada Makanan Gorengan

**Abstrak**

Kemangi adalah salah satu tanaman obat di Indonesia yang secara empiris telah digunakan sebagai antimikroba, analgetik, antiinflamasi, antivirus, antitumor, dan antioksidan yang mencegah iskemia. Tujuan penelitian ini adalah untuk mengetahui pengaruh penambahan granul ekstrak etanol daun kemangi (*Ocimum americanum*) sebagai antioksidan untuk menangkap radikal bebas pada makanan yang digoreng. Aktivitas antioksidan ekstrak etanol daun kemangi dilakukan menggunakan metode DPPH dengan penentuan $IC_{50}$. Aktivitas antioksidan dari granul dan kontrol dibandingkan dengan penentuan sifat kualitas minyak dan makanan setelah digoreng menggunakan spektra *Fourier Transfor Infra Red* (FTIR) dan parameter kuantitatif dengan penentuan persentase asam lemak bebas. Hasil maserasi daun kemangi dengan etanol didapat rendemen sebesar 15,94% b/b dengan kadar abu 9,85%, kadar abu larut air 3,98%, kadar abu tidak larut asam 3,98%, kadar ekstrak larut air 25,89%, kadar ekstrak larut etanol 12,76%, kadar air 6,87%, tingkat susut pengeringan 11,19%. Pengujian aktivitas antioksidan dari ekstrak etanol daun kemangi dengan metode DPPH diperoleh $IC_{50}$ 80,55 ppm. Hasil pengujian antioksidan dengan penentuan persentase FFA diuji secara statistik dengan ANOVA (signifikansi 0,00<5) dan perbandingan konsentrasi ekstrak dalam kemangi granul dengan uji LSD berbeda secara signifikan. Dapat disimpulkan bahwa penambahan granul ekstrak etanol daun kemangi memiliki aktivitas antioksidan pada makanan yang digoreng karena mampu menghambat peningkatan persentase FFA.

**Kata kunci**: Antioksidan, daun kemangi, DPPH, granul

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## Introduction

Globalization, urbanization, economic transition, and industrialization change the lifestyle of people. It can increase the heart disease incident. Consume unhealthy foods for examples fatty foods, baked or roasted and preserved tend to increase the risk of heart disease.[1]

Although the fatty foods consumption in Indonesia are generally at a lower level, but the cases of coronary heart disease is increase and has become one of the leading causes of death.[2] A study of the Indonesian diet showed that the main source of fat in Indonesian are fried foods (80–90%).[3] Frying is a thermal-chemical process that produces the characteristic fried food with a golden brown color, texture and flavor cracker desired appearance, so that fried food is very popular.[4]

The oil that used for frying heated continuously at high temperatures.[5] The oil will chemycally changes, the common reactions are polymerization, oxidation, hydrolysis, and produce both volatile and nonvolatile compounds.[5–6] Most of volatile compounds will be evaporated and partly through a chemical reaction and absorption in fried food products.[7]

Food products absorb the oil during the frying and cooling.[8] The oil that is absorbed during frying process is about 14–30% of total weight of the final fried food product.[7] The oil is absorbed in the food that can degrade the quality of food products. Frying causes the decline in the quality of frying oil and the quality of the fried products.[9]

The heated oil or fat used for frying on the other. The unsaturated fatty acids are unstable and the double bonds can easily transform into saturated fatty acids or trans-fatty acids that harmful to health.[10] The more the number of double bond then the more established the trans-fatty acids, especially if the oil is used repeatedly more than three times. In addition to structural changes, it will also form other compounds that are toxic.[11]

A decrease in quality of the cooking oil is causing the shelf life of the product is different from one frying process with the previous frying process.[12] Therefore, quality of cooking oil needs to be analyzed before being used again to produce a product with a shelf life that has been defined.[12,13] The quality of cooking oil is also related with security products.[12]

One effective way to prevent damage to the oil and fried products is the use of antioxidants. Synthetic antioxidants that usually use are tetra-butyl hidroquinone (TBHQ), butylated hydroxyanisole (BHA), and butylated hydroxytoluene (BHT). It is very effective for preventing oil or fat oxidation.[14] In high concentrations, TBHQ can cause cancer.[15] High concentrations and prolonged use of BHA and BHT can induce tumor. BHA can induce tumors in stomach of test animal while BHT can induce liver tumors in test animals.[16]

Many studies have showed the benefits of plants nutritious as antioxidants, such as to reduce the risk of heart disease, cancer, cataracts and other degenerative diseases due process. It makes nature antioxidants, much in demand in the world.[17] Currently the use of basil (*Ocimum americanum*) is still not optimal. One of the benefits of basil is as natural antioxidants, and it was supported by research data that showed the antioxidant activity of ethanol extract of basil leaves, namely its ability to capture free radicals DPPH with $IC_{50}$ value of 80.55 ppm.

Based on the research, granulation of ethanol extract of basil leaves determined its effect in preventing free radicals in fried foods. Research of Yoon and Choe showed that several parameters such as polar components, oxidation of free fatty acids (FFA), dienoat conjugate acid increased in each repetition frying for 60 times the period of frying.[18] Determination of free fatty acids or commonly called the FFA is crucial to the quality of the oil.[19] Because of the acid number is used to measure the amount of free fatty acids contained in the oil.[20] The larger of acid number mean that

free fatty acid content is higher.[20] It can be derived from poor processing or hydrolysis because the hydrolysis can take place with the addition of heat.[21]

On the other hand, today has developed an instrument called Fourier Transform Infra Red or FTIR spectroscopy. This instrument has advantages of being able to detect the components of a sample of food quickly and cheaply because it does not need reagents preparation. This instrument has also been applied to food widely. This is an important method in quality control and monitoring processes in food industry because of its cheap, good works, and its use is more easily compared with other methods.[21] This technique also offers a rapid and nondestructive for quantitative and qualitative analysis.[22] Therefore, FTIR spectroscopy used for detecting cooking oil damage quickly and accurately.

**Methods**

This study begins with collecting basil leaf obtained from plantations in Lembang, Bandung, West Java. The procedure of this study is divided into three stages, namely the first is preparation of the extract, the second is testing the antioxidant activity of extract, the third is preparation for making extract granule, and the last is test the quality of fried foods after adding extract granul.

In the extract preparation phase, basil previously determined in Bandungense Herbarium, Department of Biology, School of Biological Sciences and Technology ITB to know the truth of the plants then dried basil leaves using an oven at a temperature of 35 $^{o}$C and then reduced the size by using blender to obtain crude drug powder leaves basil for characteristic test include macroscopic examination, total ash determination, determination of the content of acid insoluble ash, water soluble ash content determination, water soluble ash assay, assay of soluble extract ethanol, water content determination, determination of drying shrinkage.[23,24] The next stage crude drug basil leaves was macerated with 95% ethanol then repeated 3 times each for 3 hours. The liquid extract was filtered and concentrated using rotary evaporator. The rendemen was calculated by:

$$\frac{\text{Concentrated extract} \times 100\%}{\text{Weight of crude drugs}}$$

Phytochemical screening performed that crude drugs and ethanol extracts of basil leaves covering examinate tannins, quinone, steroid and terpenoids, alkaloids, and flavonoids. Besides the monitoring of polyphenols and flavonoids class using Thin Layer Chromatography (TLC). The mobile phase varied according to the gradient polarity. Stain reagent was $FeCl_3$ 10% in methanol for detect polyphenol compounds.[25] $AlCl_3$ in methanol was stain reagent for flavonoid compounds.[26]

Antioxidant activity test for ethanol extract of basil leaf was conducted using 2,2-diphenyl-1-picrylhydrazyl or DPPH carried further by measuring the extent of the reduction reaction against DPPH free radicals can take place.[26] Measurement was made by measuring the absorbance of each sample using spectrophotometer that has been treated with a standard solution of 1 mM DPPH at λ 515 nm. The standar was ascorbic acid solution.[27]

The ethanol extract of leaves were granulated using 5% PVP, aerosil 1%, 89% lactose as a filler, the concentration of the extract used was 5%. Extract mixed with aerosil little by little until homogeneous then added PVP little by little and lactose to ethanol sufficiently homogeneous then spray until evenly mixed and can be clenched. Fist mass then sieved with 14 mesh, dried the granules by aired it. The quality of granules were evaluated includes water content test, flow rate, corner breaks and compressibility, and polyphenol and flavonoid compounds were monitored using TLC method same as that of the extract.[28] Testing the ability of ethanol extract of leaves of basil granules in preventing radical in fried foods is done by

adding the granules at a concentration of 0.1%, 0.05% and 0.025% of the total weight 100 g and then fried in oil 100 mL at 220 $^{o}$C. Negative control was made without adding granules of ethanol extract of leaves of basil. The free fatty acid (FFA) value of the samples and the control were analyzed by use titrimetry and instrumental analysis using FTIR.[29,30] The data were processed using a computer program. After that, parametric statistical test and one way ANOVA were performed followed by LSD test to determine the significance of differences in the levels of free fatty acids from four samples groups.

Sample Absorbance Spectrum Analysis with FTIR Spectroscopy was conducted to determine the spectral profile of cooking oil using FTIR spectroscopy. The FTIR used a model of IR-Prestige 21 production Shimadzu Corporation, Japan. Cooking oil samples to be measured was dripped into a KBr disk and flattened. After that, both the KBr disk were held with each other to form a sandwich KBr. Afterwards, cooking oil samples can be measured by using the wave number 400–4000 $cm^{-1}$ at resolution of 1.9. Before start the next measurement, the disk KBr should be cleaned using pure n-hexane (p.a) and wiped with a lens tissue until completely clean.[31]

Free fatty acids testing were held with titration method. Samples were stirred and then weighed as much as 5 g and put in a erlenmeyer that have been weight before and mixed 50 mL of alcohol and heated at temperature 50–75 $^{o}$C then adding 3 drops of phenolphtalein indicator to form slightly colored solution. Samples was titrated with 0.1 N NaOH. Write the NaOH volume that used to neutralize and calculate the levels of Free Fatty Acid (FFA) with the formula of calculation.[32]

## Results

Basil plant was determinated in SITH ITB based on literature indicates the plant was *Ocimum americanum* Lin. The basil leaf then dried to form crude drugs. The results of crude drug calculate. The crude drug weight was 10.54% from the weight of basil leaf. Macroscopic examination for basil leaf crude drug showed that the shape is oval bone pinnate leaf, the color is green, and the aroma is typical of basil. The crude drugs characterized and phytochemical screening was held. The result showed in Table 1 and 2.

Flavonoids and phenols in ethanol extract of basil were monitored by TLC. RF value of ethanol extract of leaves of basil use non-polar mobile fase n-hexane:ethyl acetate (7:3) was 0.775. The spotted was sprayed by spotting stain reagent $AlCl_3$ 5% in methanol for flavonoids and $FeCl_3$ 10% in methanol for phenols. Flavonoids ($AlCl_3$ 5% in methanol) showed yellow spot (+) which was confirmed by observed under 254 nm UV flourescent light spotting and polyphenols ($FeCl_3$ 10% in methanol) showed a black spot (+).[25]

The antioxidant activity test of ethanol extracts basil using DPPH method showed $IC_{50}$ values of the ethanol extract of leaf of basil was 80.55 ppm. The DPPH radical damping ability to extract ethanol mainly occurs in the secondary metabolites of polyphenols by TLC identified gave Rf

**Table 1** Characterization of Crude Basil

| No | Type of determination | Research data (w/v) |
|---|---|---|
| 1 | Levels of soluble extract ethanol | 12.76% |
| 2 | Levels of water soluble extract | 25.89% |
| 3 | Total ash | 9.85% |
| 4 | Acid insoluble ash content | 0.52% |
| 5 | Water soluble ash content | 3.98% |
| 6 | Drying shrinkage | 11.194% |
| 7 | Water content | 6.87% |
| 8 | The yield of extract | 15.94% |

**Table 2** Phytochemical Screening of Crude Basil

| No | Group of chemical compounds | Type of reagencia/examination | Result |
|---|---|---|---|
| 1 | Alcaloids | Dragendorff reagen | (-) |
| | | Mayer reagen | (-) |
| 2 | Flavonoids | Powder of Mg, (+) HCl : ethanol (1:1), and (+) amil alcohol | (+) |
| 3 | Tannins | $FeCl_3$ 1% | (+) |
| | | Gelatin 1% | (+) |
| 4 | Quinone | NaOH 1 N | (+) |
| | | Gelatin, (+) NaOH 1 N | (+) |
| 5 | Saponin | Strong shaken vertically | (+) |
| | | foam (+) HCl 2 N | (+) |
| 6 | Steroid/Triterpenoid | Lieberman-Bouchard | (+) |

value as 0.77. It was closely related to the structure of the metabolite.

The results of fried food quality test, the data analysis of free fatty acid that had undergone heating using one way ANOVA test with a value of 0.000<0.05, which indicates that there is a difference between the increase in free fatty acid groups of the samples after frying in other words, the unequal treatment of the sample affects the levels of fatty acids free on samples after fryings (Figure 1).

To find out more sample groups have the greatest effect in inhibiting the increase in free fatty acid LSD *Post Hoc* analysis.

The results of the data analysis of free fatty acid levels between the control sample (-) with three other samples 0.000<0.05 means that there was a difference between free fatty acid levels in the control sample (-) with three other samples or in other words the addition of ethanol extract granules basil leaves 0.1%, 0.05% and 0.025% effect on the levels of free fatty acids in the sample after being fried so that the increase in free fatty acid levels were not as high as in the control sample (-) (Figure 1).

## Discussion

Basil plant determination of plants in SITH ITB based literature indicates basil used was *Ocimum americanum* Lin. The basil leaf then dried to form crude drugs. The results of crude drug calculate. The crude drug weight was 10.54% from the weight of basil leaf. This shows the water content and weight of basil leaf stalks take a very large portion of which was approximately 90%.

Flavonoids and phenols in ethanol extract of basil was monitored by TLC. RF value of ethanol extract of leaves of basil use non-polar mobile fase n-hexane:ethyl acetate (7:3) was 0.775. The spotted was sprayed by spotting stain reagent $AlCl_3$ 5% in methanol for flavonoids and $FeCl_3$ 10% in methanol for phenols. Flavonoids ($AlCl_3$ 5% in methanol) showed yellow spot (+) which was confirmed by observed under 254 nm UV flourescent light spotting and polyphenols ($FeCl_3$ 10% in methanol) showed a black spot (+).[25]

Antioxidant activity test of basil leaf (*Ocimum americanum*) ethanol extract was done by a radical reduction reaction of 2,2-diphenyl-1-picrylhydrazyl or DPPH.

Free radicals were known as a major factor in the biological damage, and it used

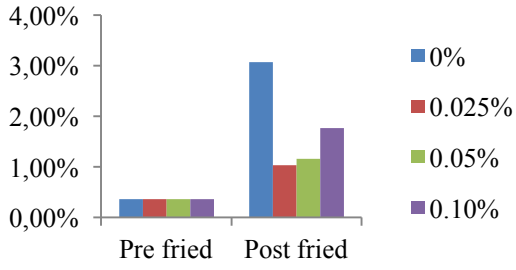


**Figure 1** Levels of Free Fatty Acid (% FFA) in Sample after Frying

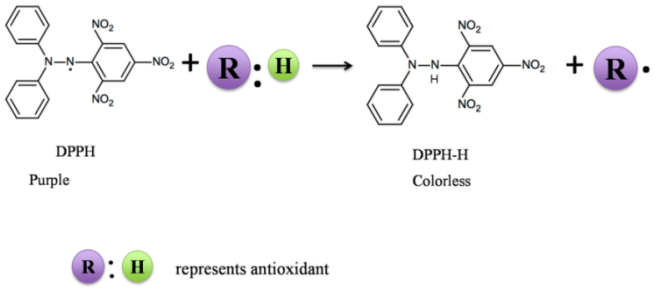


**Figure 2** Mechanism of DPPH Acceptors[33]

to evaluate the activity of DPPH free radical reduction of a natural antioxidant. The DPPH free radical was a molecule with the color purple can be transformed into a stable compound with yellow by reaction with an antioxidant, wherein antioxidant gave one electron on DPPH resulting in reduction in free radical DPPH.[34] DPPH test was a simple method test for small amount of antioxidant molecules because reaction could be observed visually by TLC, or also its intensity could be analyzed by simple spectrophotometric.[35] Structure and reaction DPPH antioxidant were showed in Figure 2.

Unpaired electrons on DPPH gives a strong absorption, maximum at λ=517 nm and purple. Suppression of free radicals by antioxidant occur when unpaired electrons become paired with a hydrogen donor, thus forming a more stable DPPH.[16] Reduction of free radical activity is usually expressed as percent inhibition of DPPH, but can also be expressed as the concentration that causes a loss of 50% DPPH activity ($IC5_0$). $IC_{50}$ values are deemed to be good measure of the efficiency of antioxidant compounds or pure extract.[18] Said to be a very strong antioxidant activity when the $IC_{50}$ values less than 50 ppm, strong if $IC_{50}$ values between 50–100 ppm, medium when the $IC_{50}$ values between 100–150 ppm, and weak when $IC_{50}$ between 151−200 ppm.[36]

$IC_{50}$ values of the ethanol extract of leaves of basil at 80.55 ppm. This means that in 80.55 ppm concentration of ethanol extract of basil leaves could reduce DPPH radical activity by 50% and classified as a strong antioxidant. DPPH radical damping ability to extract ethanol mainly occurs in the secondary metabolites of polyphenols by TLC identified gave Rf value as 0.77. It was closely related to the structure of the metabolit.

In polyphenol compounds, antioxidant activity is closely related to the structure of the side chain and also substitution on the aromatic ring.[37] Its ability to react with DPPH free radicals can affect the order of antioxidant strength. Reduction of free radical activity of polyphenolic compounds believed to be influenced by the number and position of the phenolic hydrogen in the molecule.[37] Thus the higher antioxidant activity will be generated on the amount of phenolic compounds having hydroxyl groups more at the core of flavonoids.[37]

The phenolic compounds have the ability to donate hydrogen, the antioxidant activity of phenolic compounds can be produced in the neutralization reaction of free radicals which initiate the oxidation process or the termination of radical chain reactions that occur.[23]

Antioxidant properties of flavonoids derived from the ability to transfer an electron to the free radical compounds (Figure 3).[37,38] Antioxidant also forming complexes with metal (Figure 4).[39] Both mechanisms that make flavonoids have several effects like suppress tissue damage by free radicals, inhibit lipid peroxidation and inhibit the activity of some enzymes.[40]

From the research results as well as the processing and analysis of data that has been done, it was evident that the increase in free fatty acid levels due to frying. The

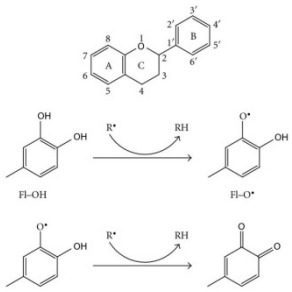

**Figure 3** The Process Suppression of Free Radicals by Flavonoids[37]

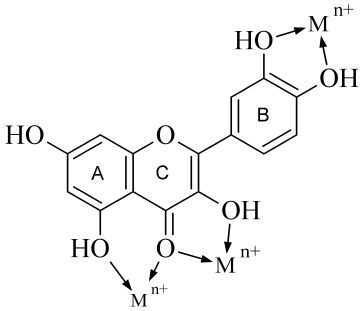

**Figure 4** Formation of Metal Complexes in Flavanoid[39]

results of data analysis of free fatty acid that had undergone heating using one way ANOVA statistic test with a value of (significance 0.000<0.05), which indicates that there was a difference between the increase in free fatty acid groups of the samples after frying in other words, the unequal treatment of the sample affects the levels of fatty acids free on samples after fryings.

Free fatty acids were the result of an overhaul that occurs in fatty acid that caused a reaction in the oil complex. This is consistent with the statement that the hydrolysis reaction that occurs in the oil will result in damage to the oil because there is a number of water in the oil and cause the formation of free fatty acids and some glycerol.[11]

To find out more sample groups have the greatest effect in inhibiting the increase in free fatty acid LSD Post Hoc analysis. The results of the data analysis of free fatty acid levels between the control sample (-) with three other samples 0.000<0.05 means that there was a difference between free fatty acid levels in the control sample (-) with three other samples or in other words the addition of ethanol extract granules basil leaves 0.1%, 0.05% and 0.025% effect on the levels of free fatty acids in the sample after being fried so that the increase in free fatty acid levels were not as high as in the control sample (-).

The addition of primary antioxidants (AH) (basil leaf ethanol extract containing polyphenolic compounds) with fatty acid low concentration may inhibit or prevent the autooxidation reaction of fats and oils.[15] The expansion may hinder the oxidation reaction stages, initiation and propagation. Radicals, antioxidants (A*) which is formed in the reaction is relatively stable and does not have enough energy react with other lipid molecules form a new fatty acid radicals.[41] The results of data analysis of free fatty acid levels between samples with addition granule of basil leaf ethanol extract 0.1% with the other two samples (0.05% and 0.025%). LSD Post Hoc test, significance value of 0.000<0.05 could be concluded that there are differences between the free fatty acid content of the samples after addition granule of basil leaf ethanol extract 0.1% with two other samples and the samples after the addition of granule extracts ethanol basil 0.05% with the sample after the addition of ethanol extract granule 0.025% basil leaves. So it could be concluded that the antioxidant ability of granules of basil leaf ethanol extract was 0.025%>0.05%>0.1% or in other words, the addition granules of basil leaf ethanol extract 0.025% has more effective antioxidant capabilities than in the addition granule of basil leaf ethanol extract 0.1% and 0.05%. It is caused by the adding granules with greater concentration of additives containing greater that could be expected to reduce the ability of ethanol extract of leaves of basil in inhibiting the increase in free fatty acids besides the more residue generated during frying which then could lead to increased levels of fatty acids be free.

## Conclusions

The results of the data analysis of free fatty acid levels between samples with the addition of granules basil leaf ethanol 0.1% with the other two samples (0.05% and 0.025%). LSD Post Hoc test, significance value of 0.000<0.05 could be concluded that there are differences between the free fatty acid content of the samples after addition granule of basil leaf ethanol extract 0.1% with two other samples and the samples after the addition of granule extracts ethanol basil 0.05% with the sample after the addition of ethanol extract granule 0.025% basil leaves. So it could be concluded that the antioxidant ability of granules of basil leaf ethanol extract was 0.025%>0.05%>0.1% or in other words, the addition granules of basil leaf ethanol extract 0.025% has more effective antioxidant capabilities than in the addition granule of basil leaf ethanol extract 0.1% and 0.05%. It is caused by the adding granules with greater concentration of additives containing greater that could be expected to reduce the ability of ethanol extract of leaves of basil in inhibiting the increase in free fatty acids besides the more residue generated during frying which then could lead to increased levels of fatty acids be free.

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## Refferences

1. Frank B. Globalization of food patterns and cardiovascular disease risk circulation. 2008;118:1913–1914.
2. Ali RAM, Suzuki T. Detection of genotoxicity of phenolic antioxidants, butylated hydroxyanisole and tert-butylhydroquinonein multiple mouse organs by the alkaline comet Assay. Life Science Journal. 2012;9(1):1.
3. Sartika RAD. Effect of trans fatty acids intake on blood lipid profile of workers in East Kalimantan, Indonesia. Malaysian Journal of Nutrition. 2011;17(1):119–127.
4. Naghshineh M. Effects of frying and storage conditions on physicochemical properties of palm olein and olive oil blends (Thesis). Serdang:Universiti Putra Malaysia;2009.
5. Aladedunyea F and Roman P. Frying stability of high oleic sunflower oils as affected by composition of tocopherol isomers and linoleic acid content. Food Chemistry. 2013;141(3): 2373–2378.
6. Seppanen CM and Saari AC. The effect of intermittent and continuous heating of soybean oil at frying temperature on the formation of 4-hydroxy-2-*trans*-nonenal and other α-, β-unsaturated hydroxyaldehydes. Journal of the American Oil Chemists' Society. 2006;83(2):121–127.
7. Choe E and Min DB. Chemistry of Deep-Fat Frying Oils. Journal of Food Science. 2007;72(5):77–86.
8. Dura´n M, Franco P, Pedro M, and Elizabeth T. Oil partition in pre-treated potato slices during frying and cooling. Journal of Food Engineering. 2007;81:257–265.
9. Gharavi N, Haggarty S, El-Kadi AOS. Chemoprotective and Carcinogenic Effects of tert-butyilhydroquinone and its metabolites. Current Drug Metabolism. 2007;8:1–7.
10. Mozaffarian D, Aro A, and Willett WC. Health effects of trans-fatty acids: experimental and observational evidence. European Journal of Clinical Nutrition. 2009;63:S5–S21.
11. Sartika RAD. Pengaruh suhu dan lama proses menggoreng (deep frying) terhadap pembentukan asam lemak

trans. Makara Sains. 2009;13(1):23–28.
12. Ngadi M, Li YS, and Sylvester O. Quality changes in chicken nuggets fried in oils with different degrees of hydrogenatation. Food Science and Technology. 2007;40(10):1784–1791.
13. Bansala G, Zhou W, Philip JB, Pranav J, Fung LN, Hui LL. Evaluation of commercially available rapid test kits for the determination of oil quality in deep-frying operations. Food Chemistry. 2010;121(2):621–626.
14. Ying Z, Lei Y, Zu YG, Chen XQ, Wang F, and Liu F. Oxidative stability of sunflower oil supplemented with carnosic acid compared with synthetic antioxidants during accelerated storage. Food Chemistry. 2010;118(3): 656–662.
15. Chaleshtori RS, N Rokni, M Rafieian K, F Drees, and E. Salehi. Antioxidant and antibacterial activity of basil (*Ocimum basilicum* L.) essential oil in beef burger. J. Agr. Sci. Tech. 2015;17:817–826.
16. Es-Safi NE, Ghidouche S, Ducrot PH. Flavonoids: hemisynthesis, reactivity, characterization and free radical scavenging activity. Molecules. 2007; 12(9):2228.
17. Lien APH, Hua H, and Chuong PH. Free radicals, antioxidants in disease and health. Int J Biomed Sci. 2008;4(2):89–96.
18. Yoon Y, Choe E. Oxidation of corn oil during frying of soy-flour-added flour dough. J Food Science. 2007;72(6): 317–23.
19. Azeman NH, Nor AY, Jaafar A, Robiah Y, Mohd NH, and Reza H. Study on the Spectrophotometric Detection of Free Fatty Acids in Palm Oil Utilizing Enzymatic Reactions. Molecules. 2015;20:12328–12340.
20. A. Fuadi Ramdja, Lisa Febrina, Daniel Krisdianto. Pemurnian Minyak Jelantah Menggunakan Ampas Tebu Sebagai Adsorben. Jurnal Teknik Kimia. 2010;17(1):7–14.
21. Maggio RM, Teodoro SK, Michele DC, Lorenzo C, Alessandra B, Angelo C, and Dario C. Monitoring of fatty acid composition in virgin olive oil by fourier transformed infra red spectroscopy coupled with partial least squares. Food Chemistry. 2009;114:1549–1554.
22. Gupta VK, Kumria R, Garg M, Gupta M. Recent updates on free radical scavenging flavonoids: an overview. Asian Journal of Sciences. 2010;9(3):108.
23. Muhtadi M. Aktivitas Antihiperurisemia Ekstrak Air Jinten Hitam (Coleus Ambonicus Lour) pada Mencit Jantan Galur Balb-C dan Standardisasinya. *Majalah Farmasi Indonesia.* 2011; 22(2):77–84.
24. Manoi F. Pengaruh Cara Pengeringan Terhadap Mutu Simplisia Sambiloto. Buletin Penelitian Tanaman Rempah dan Obat. 2006;17(1):1–5.
25. Gowri1 SS and Vasantha K. Phytochemical screening and antibacterial activity of *Syzygium cumini* (L.) (Myrtaceae) leaves extracts. International Journal of PharmTech Research. 2010;2(2): 1569–1573.
26. Ramamoorthy PK and Awang B. Antioxidant Activity, Total Phenolic And Flavonoid Content Of Morinda Citrifolia Fruit Extracts From Various Extraction Processes. Journal of Engineering Science and Technology. 2007;2(1):70–80.
27. Ozgen M, Neil RR, Artemio Z, Joseph CS, and Raymond MA. Modified 2,2-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS) method to measure antioxidant capacity of selected small fruits and comparison to Ferric Reducing Antioxidant Power (FRAP) and 2,2′-Diphenyl-1-picrylhydrazyl (DPPH) methods. J. Agric. Food Chem. 2006;54: 1151−1157.

28. Hana N. Formulasi tablet hisap ekstrak etanol gambir (*Uncaria gambir* Roxb.) dengan variasi konsentrasi polyvinyl pyrrolidone (PVP) sebagai pengikat dan pengaruhnya terhadap kadar CD4 dalam darah [diunduh 19 November 2011]. Tersedia dari: http://repository.uinjkt.ac.id/dspace/bitstream/123456789/2400/1/NAILUL%20HANA-FKIK.pdf.
29. Saad B, Cheng WL, Sariff J, Lim BP, Abdussalam SMA, Wan TW, and Idiris SM. Determination of free fatty acids in palm oil samples using non-aqueous flow injection titrimetric method. Food Chemistry**.** 2007;102 (4):1407–1414.
30. Maggio RM, Teodoro SK, Michele DC, Lorenzo C, Alessandra B, Angelo C, Dario C. Monitoring of fatty acid composition in virgin olive oil by fourier transformed infrared spectroscopy coupled with partial least squares. Food Chemistry. 2009;114:1549–1554.
31. Qing Z, Cheng L, Zhijian S, Hu XS, Qun S, and Wu J. Authentication of edible vegetable oils adulterated with used frying oil by Fourier Transform Infrared Spectroscopy. Food Chemistry. 2012;132:1607–1613.
32. Aisyah S, Eny Y, dan Ghanaim FA. Penurunan angka peroksida dan asam lemak bebas (ffa) pada proses bleaching minyak goreng bekas oleh karbon aktif polong buah kelor (*Moringa Oliefera.* Lamk) dengan aktivasi NaCl. Alchemy. 2010;1(2): 53–103.
33. Liang N and David DK. Antioxidant property of coffee components: assessment of methods that define mechanisms of action. Molecules. 2014;19(11):19180-19208.
34. Rajesh PM and Patel NJ. In vitro antioxidant activity of coumarin compounds by DPPH, Super oxide and nitric oxide free radical scavenging methods. Journal of Advanced Pharmacy Education & Research. 2011;1:52-68
35. Jothy SL, Zakaria Z and Sreenivasan S. Phytochemicals screening, DPPH free radical scavenging and xanthine oxidase inhibitiory activities of Cassia fistula seeds extract. Journal of Medicinal Plants Research. 2011;5(10):1941-1947
36. Fidrianny I, Ira R, and Komar RW. Antioxidant Capacities From Various Leaves Extracts Of Four Varieties Mangoes Using Dpph, Abts Assays And Correlation With Total Phenolic, Flavonoid, Carotenoid. International Journal of Pharmacy and Pharmaceutical Sciences. 2013;5(4):189-194.
37. Meng X, Munishkina LA, Luversky A. Effects of various flavonoids on the α-synuclein fibrillation process. Hindawi Access to Research Parkinson's Disease. 2010; Tersedia dari: http://wwwhindawi.com/journals/pd/2010/650794/
38. Calgarotto AK, Miotto S, Hono´rio KM, da Silva ABF, Marangoni S, Silva JL, Comar MJ, Oliveira KM, and da Silva SL. A multivariate study on flavonoid compounds scavenging the peroxynitrite free radical. Journal of Molecular Structure: Theochem. 2007;808:25–33
39. Ketaren S. Minyak dan lemak pangan. Jakarta: Penerbit Universitas Indonesia; 2008.
40. Boskou D, Salta FN, Chiou A, Troullidou E, Adrikopoulos NK.Conten oftrans, trans-2,4 decadienal in deep-fried and pan-fried. Journal Lipid Science Technology. 2006; 108: 109–15.
41. Laguerre M, Lecomte J, Villeneuve P. Evaluation of the ability of antioxidants to counteract lipid oxidation: Existing methods, new trends and challenges. Progress in Lipid Research. 2007;46: 244–282

## Appendix

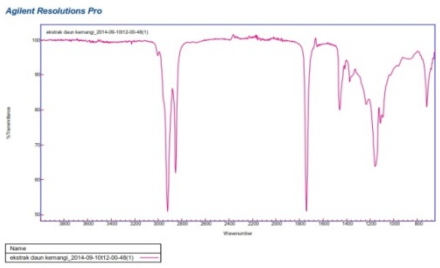


**Figure 5** FTIR Result of Basil Leaf Extract 0.1%

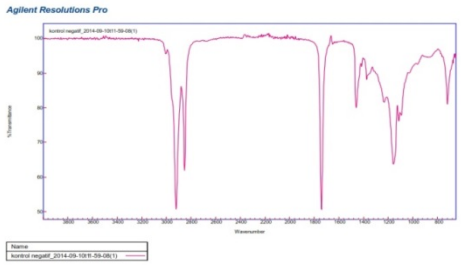


**Figure 6** FTIR Result of Negative Control the Sample was New Oil

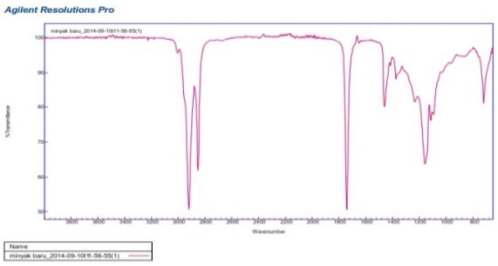


**Figure 7** FTIR Result of New Oil Sample